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Influence of Various Hydrocolloids on Suspension Stability of Chia Seeds (*Salvia hispanica* L.) in Mango Beverage and Mango Flavored Beverage

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Abstract

With the developing of juices and beverages industry, the processors need to bring new nutritional fortified products to capture the market as per the demand of the consumer who needs healthier product. Thus, this study aimed to the production of mango beverage and mango flavored beverage fortified with chia seeds; evaluate the chemical properties of chia seeds and study the effect of adding carboxymethyl cellulose, xanthan gum and low acyl gellan gum at the concentrations of 0.05% and 0.1% on the stability of suspension chia seeds in beverage during storage at ambient temperature for six months. Viscosity, color, zeta potential, suspension of seeds and organoleptic properties were evaluated. Results showed that chia seeds were rich in protein, omega-3 fatty acids and dietary fiber. The addition of 0.05% gellan gum led to improved appearance attributes and the highest stability of suspension of chia seeds for six months, and was the sample which scored highest for overall acceptability compared to the other samples of beverages. Results indicated that using chia seeds with the addition of 0.05% low acyl gellan gum led to the production of fortified mango beverage and mango flavored beverage with high stability for six months.

Keywords

Mango Beverage, Mango Flavored Beverage, Chia Seeds, Hydrocolloids, Suspension Stability

1. Introduction

In the past, beverages have been considered by the consumers only as thirstquenching and good-tasting products, while today they notably look for their health benefits [1]. These led to the development of new beverages based on different types of juices and drinks that are enriched with fruits as natural sources of nutrients, antioxidant, colors, and bioactive phytochemicals [2]. Thus, there is increasing stress on food companies to develop and produce beverages that can provide this demand, these demands and developed can be met by efficient fortification of beverage and in this status, beverage take up a distinguished place [3] [4].

Bioactive food components are utilized for enrichment of beverages it includes probiotic, phytochemicals, dietary fibers, bioactive peptides, amino acids, omega-3 fatty acids, carotenoids, antioxidants, as well as minerals and vitamins [5] [6], while integrating these fortifications into beverages, the overall stability, flavor and color, of the produced beverage are the regards of enriched beverage manufacture [5].

Mango (*Mangifera indica* L.) fruit is grown in numerous regions of the globe, especially in tropical nations, and belongs to the family of Anacardiaceae. Mango fruits are a rich source of macronutrients (proteins, fats, carbohydrates, dietary fibre) and micronutrients (vitamins, polyphenols, carotenoids, other phytochemical) which are vital to normal health and growth of human and promote health-stimulating effects, immune system, oxidative stress, gene expression in cell proliferation and apoptosis, hormone metabolism and others [7].

Chia (*Salvia hispanica* L.) is an annual herbaceous plant; seeds of these plants have been eaten for thousands of years. Nowadays, there is a growing interest and circulation of chia seeds for their nutritional composition and functional properties, which health promotes and are used in the preparation of various novel food products such as bakery products, pudding, salads. Chia seeds are a rich source of omega-3 fatty acids, protein, dietary fibers, antioxidant compounds, minerals which health benefits such as protection from inflammation, support bone health, have potential skin benefits, reduce the level of cholesterol, aging and cancer, reducing inflammation and regulating bowel function [8].

Hydrocolloids agents, xanthan gum and gellan gum are hetero polysaccharides microbially produced and by *Xanthomonas campestris* [9] and *Sphingomonas paucimobilis* [10] respectively, while the carboxymethyl cellulose is an anionic polysaccharide and man-made modified cellulose which is prepared by the reaction of monochloroacetic acid with alkali cellulose [11]. These hydrocolloids are suitable and common stabilizers due to their properties including high viscosity at low concentration and good suspension characteristics [12].

To meet the requirements and desires of various ranks of consumers towards non-traditional and healthier fortified products. Therefore, the present study was aimed to produce of mango beverage and mango flavored beverage, fortified with chia seeds; evaluate the chemical properties of chia seeds; effect of adding carboxymethyl cellulose, xanthan gum and low acyl gellan gum on the stability of suspension chia seeds in beverage during storage at ambient temperature for six months.

2. Materials and Methods

2.1. Materials

Chia (*Salvia Hispanica* L.) seeds (CHS), mango fruit (*Mangifera indica* L.) cv. Sukari, and sugar were obtained from a local market in Alexandria, Egypt. Ascorbic acid, citric acid, natural beta carotene, mango flavor, carboxymethyl cellulose (CMC), xanthan gum (XG) and low acyl gellan gum (GG) were of food-grade and obtained from Misr Food Additives (MIFAD). All chemicals and reagents were of analytical grade and purchased from El-Gomhouria Co. for Chemical and Medical Appliances, Alexandria, Egypt. Glass bottles were purchased from the local market, Alexandria, Egypt.

2.2. Technological Methods

2.2.1. Preparation of Chia Seeds (CHS)

Dry CHS were cleaned from impurities and odd materials. Dry cleaned seeds (10 g/1L beverage) were soaked in sufficient quantity of distilled water, then left for 30 min with frequent stirring until inducing full water absorption at ambient temperature (25˚C ± 5˚C).

2.2.2. Preparation of Carboxymethyl Cellulose (CMC) and Xanthan Gum (XG) Solutions

CMC and XG solutions were prepared by dissolving 0.05 and 0.1 g each from CMC and XG in 100 ml distilled water and were stirred with a magnetic stirrer for 10 min and till it was completely dissolved at ambient temperature.

2.2.3. Preparation of Gellan Gum (GG) Solution

GG solution was prepared by dissolving (0.05 g and 0.1 g) in 100 ml hot distilled water at 85˚C and adding 0.05 g calcium chloride to each solution then it was continuously stirred with a magnetic stirrer for 5 min until it was completely dissolved at 85˚C.

2.2.4. Preparation of Beverages

The preparation and processing operations of mango beverage (MB) and mango flavored beverage (MFB) were conducted in the laboratory of Edfina Company for preserved foods, according to the Egyptian standard (No. 1602/2017) as follow:

1) ***Preparation of mango beverage (MB)***

Mango fruits were carefully washed using tap water and were well drained. The skin and seeds were removed manually, then the pulp was cut into small pieces. These pieces were blended with a little water by a blender (Kenwood major blender, Japan) to obtain the mango puree (total soluble solids (TSS) 15˚Brix). Mango puree was packed in plastic bags, (each bag weighs 1 kg.), then stored at −18˚C until further use. The basic formula to produce 1 L of MB, was 100 g mango puree which represented (10%), 105 g sugar, 1.5 g citric acid, 0.25 g ascorbic acid and 783 ml water.

2) ***Preparation of mango flavored beverage (MFB)***

The basic formula of MFB was 120 g sugar, 1.5 g citric acid, 0.25 g ascorbic acid, 0.15 g beta carotene as natural coloring agent, 1.5 g mango flavor and 867 ml water, then the ingredients were well mixed.

2.2.5. Preliminary Experiments

Various preliminary experiments (sixteen samples) were performed to select the best concentrations of hydrocolloids used in this investigation, that make high suspension stability of CHS distributed in their beverages during storage for 48 h at ambient temperature, (25˚C ± 5˚C). The samples were prepared as follows:

1) One sample of MB and one sample of MFB were prepared as described by the Egyptian standard.

2) One sample of MB + CHS was prepared mango beverage-chia seeds (MB-CHS) and considered as control sample (C_1).

3) One sample of MFB + CHS was prepared mango flavored beverage-chia seeds (MFB-CHS) and considered as control sample (C_2).

4) Two samples of MB-CHS + 0.05% or 0.1% CMC.

5) Two samples of (MFB-CHS) + 0.05% or 0.1% CMC.

6) Two sample of MB-CHS + 0.05% or 0.1% XG.

7) Two sample for MFB-CHS + 0.05% or 0.1% XG.

8) Two sample for MB-CHS + 0.05% or 0.1% GG.

9) Two sample for MFB-CHS + 0.05% or 0.1% GG.

The components were mixed and stirred well at ambient temperature for uniformity, and TSS of all beverage samples were set at 12˚Brix by using a refractometer. All beverage samples were heated to 90˚C, then the hot solutions were placed in sterilized glass bottles and sealed. The glass bottles were kept at 85˚C for 10 min in a water bath, then cooled to room temperature in an icy water bath. After 48 h of storage period at ambient temperature, viscosity, color and sensory attributes were measured.

2.2.6. Basic Experiments

According to the results obtained from the preliminary experiments, the best-chosen treatments were: MB-CHS with 0.05% GG, 0.1% XG and 0.1% CMC; MFB-CHS with 0.05% GG; compared to the control samples C_1 and C_2.

All processing operations relied on the implementation of hygienic requirements. The chosen samples were packed into 200 ml of sterilized clear glass bottles, then heat treatment at 90˚C for 10 min in a hot water bath, tightly closed, then directly cooled by icy water bath. Samples were kept at ambient temperature (25˚C ± 5˚C) for six months storage period.

2.3. Physical and Chemical Analysis

2.3.1. Chemical Analyses

CHS were ground in a grain mill (Kenwood multi mill, Japan) and sieved to pass through 60 mesh sieves, then packed in glass jars and stored at 4˚C until analysis. Total solids, ash and crude fiber contents were determined according to [13]. The analyses of CHS, MB, MFB, MB-CHS with 0.05% GG and MFB-CHS with

0.05% GG were carried out as follows: crude protein, crude oil and total dietary fiber contents were determined according to the method of [13]. Carbohydrate was calculated by difference. Caloric value was calculated using the universally acceptable conversion factors by multiplying protein and carbohydrates by 4.00 and fat by 9.00 Kcal/g. Potassium was determined using flame photometer (Model PEP7, England), while calcium, magnesium, iron, zinc, manganese and copper were determined using Perkin Elmer Atomic Absorption Spectrophotometer Model 2380 [14]. The fatty acid composition of CHS was investigated by Gas Liquid Chromatography (GLC). The chia oil was esterified before GLC analysis using the method described by [15]. The chromatogram of the authentic fatty acids was used to characterize the fatty acids according to their retention times. Fatty acid composition was expressed as a percentage of the total fatty acids [16].

2.3.2. Viscosity and Color Measurement

Viscosity was measured with a rotary viscometer (Brookfield Model DV-II + Pro, USA) at the speed of 60 rpm and spindle number 2 [17]. The color attributes of MB-CHS and MFB-CHS were measured using a colorimeter (Hunter L^* a^*, b^*, Ultra scan vis, USA) with a tristimulus absorption filter. The tristimulus coordinates: L^* (lightness), a^* (redness), and b^* (yellowness) were measured [18].

2.3.3. Zeta Potential

CHS were separated from beverages and the filtrate was filled in cuvettes in order to measure the zeta potential using a zetasizer (Zetasizer Zen3600, Malvern Instruments Ltd, UK) according to [19].

2.3.4. Measurement of Seeds Sedimentation

To measure the stability of CHS in beverage height percentage of seeds was visually measured during 48 h and six months of storage compared to the control samples (C_1 and C_2) using the following equation [19].

$$\text{Height percentage of seeds}(\%) = \frac{\text{Height of seeds}}{\text{Total sample height in the bottle}} \times 100$$

The best-chosen samples depended on the results of suspension stability of CHS after 48 h of storage.

2.3.5. Sensory Evaluation

The tests of sensory parameters in the preliminary and basic experiments were conducted using a 9-point hedonic scale [20]. The judgment was accomplished by 20 panelists, including 10 panelists from Food Technology Laboratory, of Sabahia, Alexandria, Egypt, Food Technology research institute (FTRI) and 10 panelists from Edfina company, Alexandria, Egypt. Samples were presented to the panelists in 250 ml clear bottles, that were randomly coded. The sensory attributes evaluated were color, taste, texture, mouthfeel, appearance & suspension of seeds and overall acceptability.

2.3.6. Statistical Analysis

All analysis were triplicate, except for fatty acids, colorimetric measurements and zeta potential measurements, and were expressed as mean values ± standard deviation. Statistical analyses were carried out using software (SAS) version 9.1 (SAS Institute Inc., Cary, NC) [21]. Statistical analyses were performed using one-way analyses of variance followed by Duncan's test. Differences were considered significant at $p \leq 0.05$.

3. Results and Discussions

3.1. Chemical Analyses of CHS

Table 1 illustrates the proximate composition of CHS. Protein, lipid, and carbohydrates recorded 23.50%, 35.28% and 4.54%, respectively. Ash, crude fiber, and total dietary fiber gave values of 14.96%, 21.73% and 32.63%, respectively. These results indicated that lipid is the most abundant component in CHS and provide 45.23% of daily value (DV), followed by total dietary fiber providing an excessive amount of 116.54% DV and protein that was of 47% DV. As a result, CHS can be considered as a new oil source and is also a good source of plant protein and dietary fiber. These outcomes were consistent with [22] who mentioned that CHS contained high levels of fiber and that a high amount of fiber decreases the risk of coronary heart disease, the risk for diabetes type 2 and several types of cancer. These results were also in agreement with [23].

Table 1 also shows that CHS constitutes a source of minerals. CHS had a high level of macroelements including calcium 562.52, potassium 467.5 and magnesium 337.56 mg/100g. Microelements; iron 7.35, zinc 4.95, manganese 2.55, and copper 0.97 μg/100g). [24] found that the concentration of macroelements in chia is as; calcium 631, potassium 407, magnesium 335 mg/100g. Microelements; copper 0.924, iron 7.72, manganese 2.72 and zinc 4.58 μg/100g. These results show that CHS provided 43.27% DV of calcium, 80.37% DV of magnesium.

The fractions of fatty acid in CHS oil were shown in **Table 1**. The saturated and unsaturated fatty acids were 9.49% and 89.94%, respectively. Three dietary and healthy unsaturated fatty acids were identified as follows: $\omega 3$ linolenic acid (65.84%), it was the major component of fatty acid, followed by $\omega 6$ linoleic acid (17.10%) and ω-9 oleic acid (7%). Otherwise, the saturated fatty acids included palmitic (6.85%), stearic (2.42%), and lauric acid (0.22%). The ratio between saturated and unsaturated fatty acids was (1:9.48) and was (1:3.85) for $\omega 6$:$\omega 3$ fatty acids. These results were in agreement with [22] who mentioned that CHS is a rich source $\omega 3$ fatty acids, that improve cognitive performance, protect from inflammation and lower the cholesterol. Additionally, [25] who stated that the CHS oil have a high concentration of α-linolenic acid, which is used in functional foods preparation. CHS oil has a high amount of α-linolenic acid, which is crucial for protecting the heart and liver [26].

Depending on the for mentioned results, the CHS are considered a good source of ω-3 fatty acids, and dietary fiber, thus could be used for fortifying mango beverage and flavored mango beverage prepared in these investigation.

Table 1. Chemical composition of chia (*Salvia hispanica* L.) seeds (on dry weight basis).

Components	Mean value	% DV
<u>Proximate composition (g/100g)</u>		
Total solids	92.85 ± 0.40	--
Ash	14.96 ± 0.57	--
Protein	23.50 ± 0.72	47
Lipid	35.28 ± 0.86	45.23
Crude fiber	21.73 ± 0.66	77.60
Carbohydrate	4.54 ± 0.43	1.65
Total dietary fiber	32.63 ± 0.92	116.54
Energy value (Kcal/100g)	471.32 ± 5.86	
<u>Mineral</u>		
<u>Macroelements</u> (mg/100g)		
Calcium	562.52	43.27
Potassium	467.50	9.95
Magnesium	337.56	80.37
<u>Microelements</u> (µg/100g)		
Iron	7.35	-
Zinc	4.95	-
Manganese	2.55	-
Copper	0.97	-
<u>Fatty acid (%)</u>		
<u>Saturated</u>		
Lauric acid (C12:0)	0.22	--
Palmitic acid (16:00)	6.85	--
Stearic acid (C18:0)	2.42	--
Total of saturated fatty acid	**9.49**	
<u>Unsaturated</u>		
<u>1) Monounsaturated</u>		
Oleic (C18:1, *ω*-9)	7.00	--
<u>2) Polyunsaturated</u>		
Linoleic acid (C18:2, *ω*-6)	17.10	--
Linolenic acid C18:3, *ω*-3)	65.84	--
Total of unsaturated fatty acid	**89.94**	--
<u>Sat: Unsat ratio</u>	1:9.48	--
*ω*6: *ω*3 FA ratio	1:3.85	--

Data represented the means ± standard deviation; *ω*-3: omega 3; *ω*-6: omega 6; *ω*-9: omega 9; % DV: % Daily Value.

3.2. The Parameters Used in the Preliminary Experiments

3.2.1. Viscosity

The effect of hydrocolloid type and its concentration on the viscosity of studied beverages, after 48 h of storage at ambient temperature, as preliminary experiments were shown in Figure 1. It could be observed that the lowest viscosity was in C_2 (26 cps) followed by C_1, which recorded (28 cps). The low viscosity is due to the absence of hydrocolloid, Figure 1. The samples of MB-CHS recorded higher viscosity than MFB-CHS as compared with samples containing the same

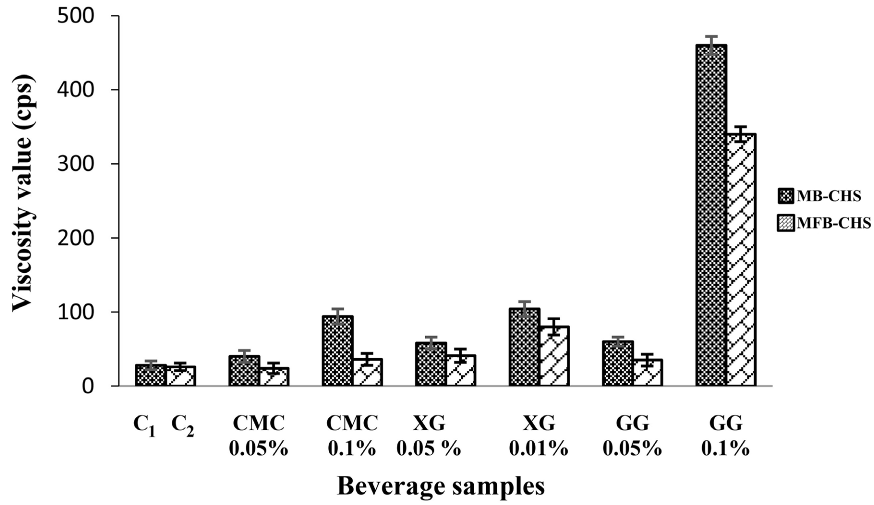

Figure 1. Effect of different types and concentrations of hydrocolloids on the viscosity of the beverages after storage at ambient temperature for 48 h MB-CHS: mango beverage-chia seeds; MFB-CHS: mango flavored beverage-chia seeds; CMC: carboxymethyl cellulose; XG: xanthan gum; GG: low acyl gellan gum; C_1: mango beverage without gum; C_2: mango flavored beverage without gum.

type and concentration of hydrocolloids. This difference may be due to using natural mango pulp for processing of MB-CHS. The addition of CMC, XG and GG at the concentration of (0.05% and 0.1%) in MB-CHS and MFB-CHS samples, led to significantly higher viscosity ($P \leq 0.05$) as compared with C_1 and C_2, respectively. This means that, increase of hydrocolloid concentration led to the increase of beverage viscosity. These results agree with those by found of [27] who mentioned that increasing the concentration of gum in drinks caused a direct proportional increase in the viscosity and sensory perception. The highest viscosity was observed in MB-CHS and MFB-CHS containing GG 0.1%, they were (460 cps and 340 cps), respectively. The high viscosity led to formation of hard gel of the studied beverages. These results were in agreement with [28] who stated that if the concentration of the gellan gum is sufficiently high, then the double helices form can further transform into thicker rod-like aggregates leading to the formation of a macroscopic gel.

3.2.2. Suspension Stability of CHS

The influence of using CMC, XG and GG at the concentration of 0.05% and 0.1% compared with the control sample on the stability of suspension of CHS in MB and MFB during storage 48 h is illustrated in **Figure 2(a)** and **Figure 2(b)**. C_1 and C_2 as control samples recorded the lowest percentage of stability and suspension, while the samples containing CMC and XG at the concentration of 0.05% gave a weak stability. The fast rate of seeds sedimentation was observed in the sample of MFB-CHS during the first hour of storage, and the best stability and suspension of seeds was observed in MB-CHS; these results were due to using the natural pulp of mango fruit in the processing of MB. In both MB-CHS and MFB-CHS beverages, the addition of GG (0.05% and 0.1%) led to the increase

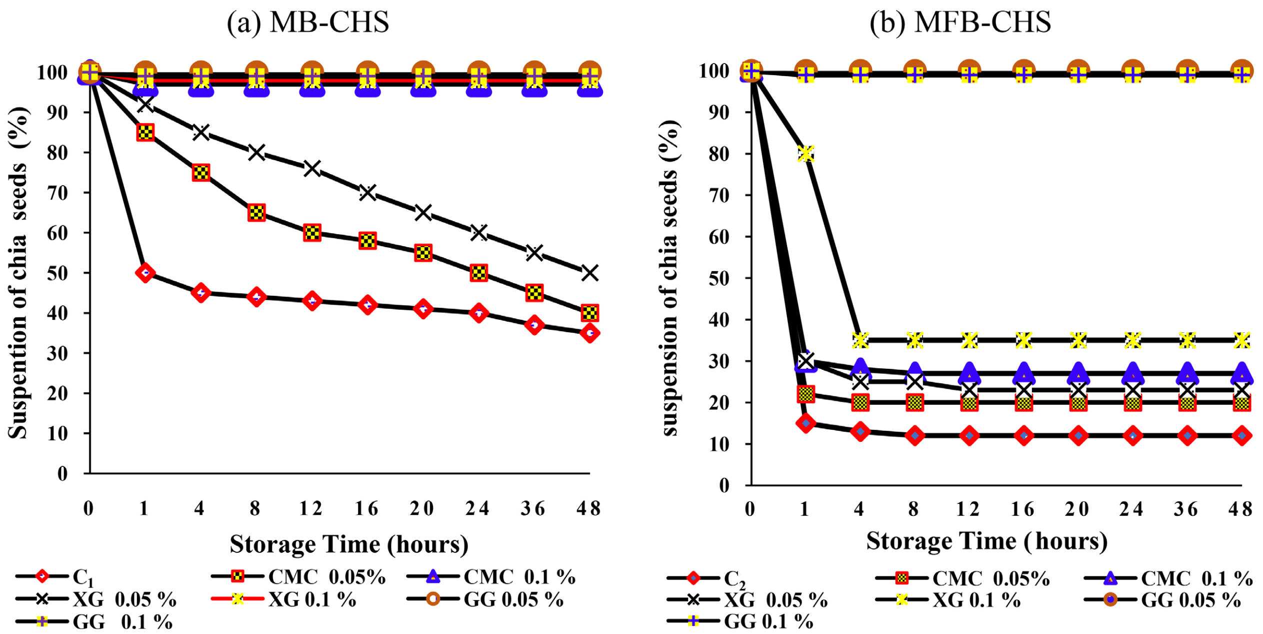

Figure 2. Effects of different types and concentrations of hydrocolloids on the suspension stability of chia seeds in beverage during storage for 48 h at ambient temeprature. MB-CHS: mango beverage-chia seeds; MFB-CHS mango flavored beverage-chia seeds; CMC: carboxymethyl cellulose; XG: xanthan gum; GG: low acyl gellan gum; C_1: mango beverage without gum; C_2: mango flavored beverage without gum.

stability and suspension of seeds, also the addition of 0.1% XG and 0.1% CMC in MB-CHS gave the high stability and suspension of seeds. While no effect was noticed on the stability of MFB-CHS. No significant difference in stability was observed between the three hydrocolloids when added to MB-CHS. These results agree with [29] who reported that XG is effective in suspending fruit pulp for long periods. [30] mentioned that GG fluid gels can be used to produce shelf stable suspensions in a variety of beverages products.

3.2.3. Color

The effect of different types and concentrations of hydrocolloids on color values (L^*, a^* and b^*) of beverages after processing in storage for 48 h at 25˚C ± 5˚C are given in Table 2. A slight decrease was shown in the L^* value in all treatments compared with controls (C_1 and C_2), while, this decrease of these values was higher by adding XG and GG compared with the sample containing CMC. This means that addition of XG and GG, led to color stable of beverages. These results agree with [31] and [32], who found that the presence of GG and XG had increased the stability of acerola smoothie and red rice drinks pigments such as anthocyanins and carotenoids when compared to the control. For the values a^* and b^*, all treatments have higher values than the control, however, change the concentrations from 0.05% to 0.1% of hydrocolloids had not effect on L^*, a^* and b^* values in all the studied samples, these results are in agreement with [33].

3.2.4. Sensory evaluation

After storage at ambient temperature for 48 h the average scores for the parameters

Table 2. Effect of type and concentration of hydrocolloids on color parameters of mango beverage and mango flavored beverage fortified with chia seeds after 48 h of processing.

Sample	Concentration (%)	CIE Color values		
		L^*	a^*	b^*
MB-CHS				
C_1	0.00	35.78	4.35	12.54
CMC	0.05	35.31	5.32	16.71
	0.10	35.27	5.38	16.78
XG	0.05	34.91	4.88	16.19
	0.10	34.98	4.93	16.23
GG	0.05	34.46	4.90	17.13
	0.10	34.66	4.52	17.06
MFB-CHS				
C_2	0.00	38.94	12.40	20.22
CMC	0.05	36.63	14.08	24.92
	0.1	36.31	14.96	24.11
XG	0.05	35.24	13.29	23.31
	0.1	35.28	13.48	23.27
GG	0.05	35.81	13.06	22.93
	0.1	35.97	13.33	22.88

L^*: lightness; a^*: redness; b^*: yellowness; MB-CHS: mango beverage-chia seeds; MFB-CHS: mango flavored beverage-chia seeds; CMC: carboxymethyl cellulose; XG: xanthan gum; GG: low acyl gellan gum; C_1: mango beverage without gum; C_2; mango flavored beverage without gum.

of sensory attributes of MB-CHS and MFB-CHS containing hydrocolloids at concentration of 0.05% and 0.1% are presented in Table 3. There were no significant differences ($p \geq 0.05$) among treatments for color and taste after 48 h of processing. The texture and mouthfeel satisfaction increased with the addition of 0.05% GG, 0.1% CMC and 0.1% XG, while there was significant decrease ($p \leq 0.05$) of these properties when 0.1% GG was added to MB-CHS and MFB-CHS, and that was reflected in the decision of panelists, who disliked the samples with 0.1% GG. These results were confirmed with viscosity analyses Figure 1. With increasing CMC and XG concentration in MB-CHS, the appearance and suspension of seeds increased ($p \leq 0.05$). On the other hand, 0.05%, 0.1% GG gave highest scores and were extremely liked concerning appearance and suspension of seeds in MB-CHS and MFB-CHS. The overall acceptability recorded the highest score for the treatment containing 0.05% GG of both beverages followed by the treatments containing 0.1% XG and 0.1% CMC when added to MB-CHS sample. These results agree with [29] and [28], they reported that XG and GG imparted a drink-enhanced mouth feel with a full-bodied taste and good flavor release.

Table 3. Average scores for the sensory attributes of mango beverage and mango flavored beverage fortified with chia seeds after 48 h of processing.

Sensory property		Color	Taste	Texture	Mouthfeel	Appearance & Suspension of seeds	Overall acceptability
Treatment	Con (%)	MB-CHS					
C_1	0.00	8.29 ± 0.49^a	8.35 ± 0.53^a	7.43 ± 1.13^b	7.53 ± 0.56^b	2.71 ± 0.76^b	4.30 ± 0.82^b
CMC	0.05	8.36 ± 0.53^a	8.42 ± 0.53^a	7.57 ± 0.98^b	7.64 ± 0.75^b	2.79 ± 0.57^b	4.44 ± 0.70^b
	0.10	8.43 ± 0.78^a	8.40 ± 0.59^a	8.71 ± 0.36^a	8.49 ± 0.69^a	8.26 ± 0.72^a	8.50 ± 0.48^a
XG	0.05	8.67 ± 0.63^a	8.42 ± 0.75^a	7.50 ± 1.04^b	7.57 ± 0.53^b	2.71 ± 1.11^b	4.59 ± 1.11^b
	0.10	8.71 ± 1.07^a	8.43 ± 0.70^a	8.72 ± 0.49^a	8.50 ± 0.69^a	8.36 ± 0.73^a	8.49 ± 0.38^a
GG	0.05	8.79 ± 0.40^a	8.50 ± 0.50^a	8.79 ± 0.39^a	8.79 ± 0.37^a	9.00 ± 0.0^a	8.80 ± 0.38^a
	0.10	8.79 ± 0.27^a	8.51 ± 0.71^a	4.00 ± 0.82^c	4.43 ± 1.27^c	9.00 ± 0.0^a	4.59 ± 0.86^b
		MFB-CHS					
C_2	0.00	8.20 ± 0.63^A	8.40 ± 0.50^A	5.57 ± 0.53^C	6.70 ± 0.54^C	2.43 ± 0.79^C	4.10 ± 0.79^B
CMC	0.05	8.16 ± 0.75^A	8.21 ± 0.70^A	7.00 ± 0.82^B	7.50 ± 0.50^B	2.5 ± 0.65^C	4.36 ± 0.70^B
	0.10	8.14 ± 0.70^A	8.26 ± 0.62^A	8.51 ± 0.38^A	8.53 ± 0.62^A	3.10 ± 0.58^B	4.53 ± 0.73^B
XG	0.05	8.19 ± 0.78^A	8.21 ± 0.75^A	7.18 ± 0.60^B	7.14 ± 0.70^{BC}	2.50 ± 0.50^C	4.40 ± 0.81^B
	0.10	8.18 ± 0.73^A	8.25 ± 0.40^A	8.43 ± 0.53^A	8.34 ± 0.66^A	3.14 ± 0.69^B	4.50 ± 0.58^B
GG	0.05	8.19 ± 0.54^A	8.32 ± 0.31^A	8.71 ± 0.39^A	8.73 ± 0.36^A	9.00 ± 0.0^A	8.79 ± 0.19^A
	0.10	8.19 ± 0.60^A	8.30 ± 0.50^A	4.14 ± 0.90^D	4.00 ± 1.00^D	9.00 ± 0.0^A	4.73 ± 0.53^B

MB-CHS: mango beverage-chia seeds; MFB-CHS: mango flavored beverage-chia seeds; CMC: carboxymethyl cellulose; XG: xanthan gum; GG: low acyl gellan gum; C_1: mango beverage without gum, C_2: mango flavored beverage without gum; Different superscripts in the same column are significant differences at P ≤ 0.05 level.

From the previous results of preliminary experiments, the best treatments were chosen according to the resulted of stability of suspension CHS, non-sedimentation, color values, uniformity as well as the organoleptic properties of studied beverages. This was an indicator to conduct the basic experiments and the chosen treatment were as follows: CHS with (0.05% GG, 0.1% XG and 0.1% CMC) for preparing MB-CHS and CHS with 0.05% GG for preparing MFB-CHS and then studying their behavior during storage for six-months at ambient temperature (25˚C ± 5˚C).

3.3. Basic Experiments

3.3.1. Suspension Stability of Seeds during Storage

The effect of the gum types on the suspension of CHS in MB and MFB during the storage period of six months are shown in Figure 3(a) and Figure 3(b). All beverages recorded high stability of suspension of CHS in the initial time of storage Figure 2(a) and Figure 2(b). The control samples (C_1 and C_2) recorded low stability of suspension of seeds and fast rate of seeds sedimentation.

During storage period of six months, the beverage containing 0.05% GG in MB-CHS (Figure 3(a)) and MFB-CHS (Figure 3(b)) recorded high stability of suspension of CHS, followed by 0.1% XG in MB-CHS, which recorded high stability of suspension of seeds at the initial two months of storage, and showed a

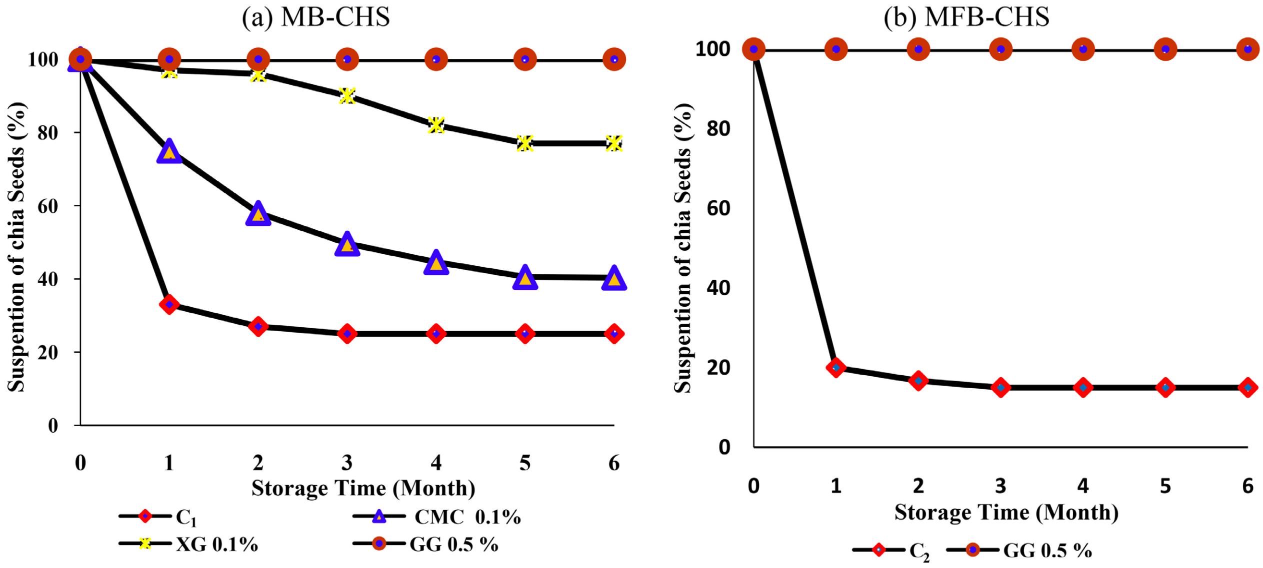

Figure 3. Effects of different gum types on suspension stability of chia seeds in beverages during storage for six months at ambient temperature. MB-CHS: mango beverage-chia seeds; MFB-CHS: mango flavored beverage-chia seeds; CMC: carboxymethyl cellulose; XG: xanthan gum; GG: low acyl gellan gum; C_1: mango beverage without gum; C_2: mango flavored beverage without gum.

significant decrease ($P \leq 0.05$) by the increase of storage period for six months. During the storage period, the suspension of seeds in beverages containing CMC 0.1% significantly decreased ($P \leq 0.05$) by increasing storage time as compared with 0.05% GG and 0.1% XG. These results showed that the use of 0.05% GG led to highest stability and the best appearance of suspension of CHS during six months of storage (visual observation), as they almost fully retained their physical properties during storage. These results agreed with those reported by [34]

3.3.2. Zeta Potential

Dispersed systems such as colloidal suspensions contain electrically charged particles that interact with each other and with the media, these amounts of charges are called zeta potential, and are used to gauge how stable is colloidal system [35]. As shown in **Figure 4**, the zeta potential values for all samples were clearly negative, indicating that there are more negatively charged particles than positively charged particles in all beverages; this was attributed to the existence of beverage particles with positively charged carbohydrate and protein nuclei surrounded by negatively charged pectin (C_1 and C_2) as well as to the properties of added stabilizers to the samples [33]. Furthermore, the absolute value of zeta potential of the beverage samples with XG, CMC or GG were higher than that of the control (C_1; −14 and C_2; −12 MV). This is because the XG, CMC and GG are anionic polysaccharides and thus provide a certain amount of negatively charged particles, [32] [36]. In the initial time, the sample containing 0.1% XG recorded higher value of zeta potential (−27.1 MV) followed by 0.1% CMC (−22.2 MV), while the sample containing 0.05% GG recorded the lowest value (−21.4 MV in MB-CHS and −21 MV in MFB-CHS). These results may be due to adding double

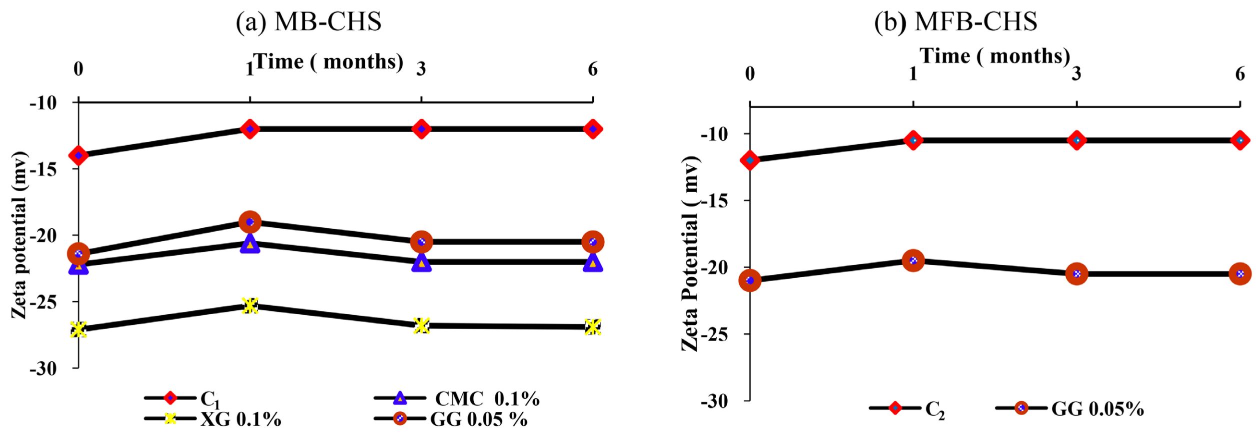

Figure 4. Effect of different gum types on zeta potential of beverages fortified with chia seeds during storage for six months at ambient temeprature. MB-CHS; mango beverage-chia seeds; MFB-CHS: mango flavored beverage-chia seeds; CMC: carboxymethyl cellulose; XG: xanthan gum; GG: low acyl gellan gum; C_1: mango beverage without gum, C_2: mango flovored beverage without gum.

concentration of XG and CMC compared with GG. These results agreed with [37] and [33] who mentioned that the absolute zeta increased with the increase in hydrocolloid concentration due to the anionic properties of hydrocolloids. During the storage period for one-month, absolute zeta slightly decreased in all samples. This change in zeta may be due to the exit of protein with positive charge from CHS [38]. Zeta value in C_1 and C_2 decreased after one month then become stable tell six months of storage, while in beverage with different gum types, zeta potential showed a decreased after one month then increased after three months, these may be due to the anionic and adsorbing properties of the existing hydrocolloids (XG, CMC and GG). Finally, is become stable tell the end of storage for six months. Similar outcomes have been attained when pectin, gum tragacanth, CMC, XG and GG were used as stabilizers in juices and dairy products [37] and [39]. Although the samples of 0.05% GG recorded lower zeta potential but it had the highest stability of seeds suspension, these results agree with [40] who mentioned that, it is common to find stable systems with low absolute zeta potential values and vice versa.

3.3.3. Sensory Attributes after Six Months Storage Period

The sensory attributes of the selected beverages after storage for six months are illustrated in **Figure 5(a)** and **Figure 5(b)**. Generally, the beverages containing 0.05% GG in MB-CHS and MFB-CHS recorded the highest mean scores for all parameters compared to other treatments, was most acceptable from the organoleptic point of view and kept the suspension of seeds even at the end of six months of storage. These results confirmed by the result of seed suspension in **Figure 3**. The sensory scores of all beverage samples decreased after six months of storage period as compared to those evaluated after 48 h of storage shown in **Table 3**. The control beverage C_1 in MB-CHS and C_2 in MFB-CHS had the lowest scores of all parameters compared to the other samples and showed a decreased acceptability after six months of storage (**Table 3** and **Figure 5**). The

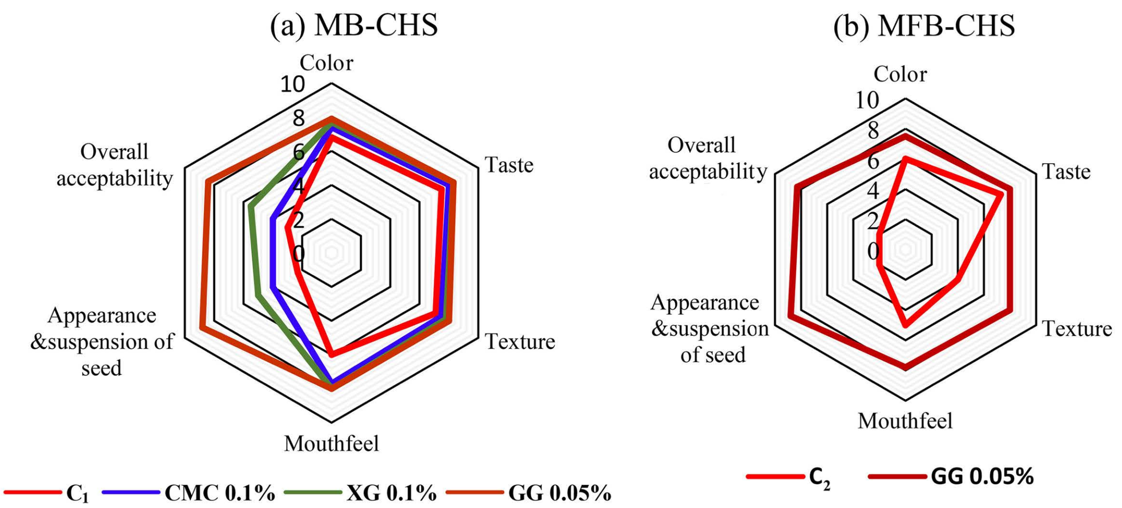

Figure 5. Average scores for the sensory attributes of beverages fortified with chia seeds after storage for six months at ambient temeprature. MB-CHS: mango beverage-chia seeds; MFB-CHS: mango flavored beverage chia seeds; CMC: carboxymethyl cellulose; XG: xanthan gum; GG: low acyl gellan gum; C_1: mango beverage without gum; C_2: mango flavored beverage without gum.

Table 4. Nutritional facts of beverages before and after fortification with chia seeds.

Component	MB		MB-CHS		MFB		MFB-CHS	
	Value	% DV	Value	% DV	Value	% DV	Value	% DV
Energy value (Kcal/L)	485.94 ± 8.17	-	547.04 ± 12.70	-	480.00 ± 10	-	541.28 ± 12.09	-
Fat (g/L)	0. 30 ± 0.01	0.38	3.80 ± 0.10	4.87	-	-	3.52 ± 0.21	4.51
Protein (g/L)	0. 81 ± 0.02	1.62	3.21 ± 0.09	6.42	-	-	2.40 ± 0.15	4.80
Carbohydrate (g/L)	120.00 ± 2.00	43.64	125 ± 3.00	45.45	120.00 ± 2.5	43.64	125.00 ± 2.4	45.45
Total dietary fiber (g/L)	1.54 ± 0.03	5.50	4.85 ± 0.12	17.32	-	-	3.30 ± 0.22	11.79
Mineral (mg/L)								
Calcium	11.00	0.85	67.23	5.17	-	-	56.30	4.33
Potassium	165.00	3.51	212.00	4.51	-	-	46.74	0.99
Magnesium	11.00	2.62	44.80	10.67	-	-	33.77	8.04

%DV: % Daily Value; MB: mango beverage; MFB: mango flavored beverage; MB-CHS: mango beverage-chia seeds; MFB-CHS: mango flavored beverage-chia seeds.

overall acceptability of the beverage containing 0.1% CMC in MB-CHS decreased with increasing storage time followed by the treatment containing 0.1% XG.

3.4. Nutritional Facts of Beverage

Nutritional facts of MB and MFB before and after fortification with CHS are shown in Table 4. The fortification of 1 L of MB and MFB with 1% CHS (MB-CHS and MFB-CHS) can meet 4.87% and 4.51% of the DV for fat, 6.42% and 4.80% for protein, 45.45% for carbohydrates finally 17.32% and 11.79% for the total dietary fiber respectively of beverage. The fortification of the beverages with CHS provided higher percentages of the daily value of different minerals compared the beverages without fortification as shown in Table 4.

5. Conclusions

From the analytical data and the previous results, it could be concluded that the fortification by chia seeds gave beverages rich in omega-3 fatty acids and dietary fiber. This is very important for human health, especially in flavored beverage without natural pulp, and meets the demand of consumers for healthy food desires. Also, using hydrocolloids in this study solved the problem of sedimentation and low suspension stability that occur in juices and beverages.

Finally, the addition of 0.05% low acyl gellan gum to the beverages led to higher stability of suspensions of CHS in the beverages and improved the appearance as compared with the addition of XG and CMC.

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Consent to Participate

All authors consent to participate (were responsible for the conceptualization, experimental design, methodology, performed formal analyses, wrote the original draft of the manuscript, revised, edited, and confirmed the manuscript).

Consent for Publication

All authors consent for publication (data or image curation and analysis).

Availability of Data and Material

All data and materials were availability.

Authors' Contributions

All authors contributed equally to this research work. R.A.A., E.A.A. and E.S.A were responsible for the conceptualization, experimental design, methodology, performed formal analyses, data curation and analysis. All authors wrote the original draft of the manuscript, revised, edited, and confirmed the manuscript.

Conflicts of Interest

We have no conflicts of interest to disclose.

References

[1] Shahidi, F. and Ambigaipalan, P. (2016) Beverages Fortified with Omega-3 Fatty Acids, Dietary Fiber, Minerals, and Vitamins. In: Shahidi, F. and Alasalvar, C., Eds., *Handbook of Functional Beverages and Human Health*, CRC Press, Boca Raton, 801-813.

[2] Gironés-Vilaplana, A., Valentão, P., Andrade, P.B., Ferreres, F., Moreno, D.A. and García-Viguera, C. (2015) Beverages of Lemon Juice and Exotic Noni and Papaya

with Potential for Anticholinergic Effects. *Food Chemistry*, **170**, 16-21. https://doi.org/10.1016/j.foodchem.2014.08.021

[3] Keenan, D.F., Brunton, N., Gormley, R. and Butler, F. (2011) Effects of Thermal and High Hydrostatic Pressure Processing and Storage on the Content of Polyphenols and Some Quality Attributes of Fruit Smoothies. *Journal of Agricultural and Food Chemistry*, **59**, 601-607. https://doi.org/10.1021/jf1035096

[4] Young, P.W., Mills, T.B. and Norton, I.T. (2021) Influence of pH on Fluid Gels Produced from Egg and Whey Protein Isolate. *Food Hydrocolloids*, **111**, Article ID: 106108. https://doi.org/10.1016/j.foodhyd.2020.106108

[5] Kasapoğlu, K.N., Daşkaya-Dikmen, C., Yavuz-Düzgün, M., Karaça, A.C. and Özçelik, B. (2019) Enrichment of Beverages with Health Beneficial Ingredients. In: Grumezescu, A.M. and Holban, A.M., Eds., *Value-Added Ingredients and Enrichments of Beverages*, Vol. 4, Academic Press, Cambridge, 63-99. https://doi.org/10.1016/B978-0-12-816687-1.00003-5

[6] Fernandes, S.S., Coelho, M.S. and de las Mercedes Salas-Mellado, M. (2019) Bioactive Compounds as Ingredients of Functional Foods: Polyphenols, Carotenoids, Peptides from Animal and Plant Sources New. In: Campos, M.R.S., Ed., *Bioactive Compounds: Health Benefits and Potential Applications*, Woodhead Publishing, Sawston, 129-142. https://doi.org/10.1016/B978-0-12-814774-0.00007-4

[7] Maldonado-Celis, M.E., Yahia, E.M., Bedoya, R., Landázuri, P., Loango, N., Aguillón, J. and Guerrero Ospina, J.C. (2019) Chemical Composition of Mango (*Mangifera indica* L.) Fruit, Nutritional and Phytochemical Compounds. *Frontiers in Plant Science*, **10**, Article 1073. https://doi.org/10.3389/fpls.2019.01073

[8] De Falco, B., Amato, M. and Lanzotti, V. (2017) Chia Seeds Products: An Overview. *Phytochemistry Reviews*, **16**, 745-760. https://doi.org/10.1007/s11101-017-9511-7

[9] García-Ochoa, F., Santos, V.E., Casas, J.A. and Gómez, E. (2000) Xanthan Gum: Production, Recovery, and Properties. *Biotechnology Advances*, **18**, 549-579. https://doi.org/10.1016/S0734-9750(00)00050-1

[10] Sutherland, I.W. (1999) Microbial Polysaccharide Products. *Biotechnology and Genetic Engineering Reviews*, **16**, 217-230. https://doi.org/10.1080/02648725.1999.10647976

[11] Sutherland, I.W. (1998) Novel and Established Applications of Microbial Polysaccharides. *Trends in Biotechnology*, **16**, 41-46. https://doi.org/10.1016/S0167-7799(97)01139-6

[12] Khan, T., Park, J.K. and Kwon, J.-H. (2007) Functional Biopolymers Produced by Biochemical Technology Considering Applications in Food Engineering. *Korean Journal of Chemical Engineering*, **24**, 816-826. https://doi.org/10.1007/s11814-007-0047-1

[13] Latimer, G.W. (2016) Official Methods of Analysis of AOAC International. 20th Edition, AOAC International, Rockville.

[14] Coorey, R., Tjoe, A. and Jayasena, V. (2014) Gelling Properties of Chia Seed and Flour. *Journal of Food Science*, **79**, E859-E866. https://doi.org/10.1111/1750-3841.12444

[15] Stahle, E. (1967) Thin Layer Chromatography. Springer, New York.

[16] Farag, R.S., Abdel Rahim, E.A., Hewedy, F.M. and Ragab, A.A. (1984) Biochemical Studies on Lipids of Hen's Egg during Incubation. *SÖFW* (*Seifen*, *Öle*, *Fette*, *Wachse*), **110**, 63-66.

[17] Mutlu, M., Sarõo Glu, K., Demir, N., Ercan, M.T. and Acar, J. (1999) The Use of Commercial Pectinase in Fruit Juice Industry. Part I: Viscosimetric Determination

of Enzyme Activity. *Journal of Food Engineering*, **41**, 147-150.
https://doi.org/10.1016/S0260-8774(99)00088-6

[18] Bursać Kovačević, D., Putnik, P., Dragović-Uzelac, V., Pedisić, S., Režek Jambrak, A. and Herceg, Z. (2016) Effects of Cold Atmospheric Gas Phase Plasma on Anthocyanins and Color in Pomegranate Juice. *Food Chemistry*, **190**, 317-323. https://doi.org/10.1016/j.foodchem.2015.05.099

[19] Genovese, D.B. and Lozano, J.E. (2001) The Effect of Hydrocolloids on the Stability and Viscosity of Cloudy Apple Juices. *Food Hydrocolloids*, **15**, 1-7. https://doi.org/10.1016/S0268-005X(00)00053-9

[20] Meilgaard, M.C., Carr, B.T. and Civille, G.V. (1999) Sensory Evaluation Techniques. CRC Press, Boca Raton. https://doi.org/10.1201/9781003040729

[21] SAS Institute Inc. (2004) SAS/ETS 9.1 User's Guide. SAS Publishing, Cary.

[22] Knez Hrnčič, M., Ivanovski, M., Cör, D. and Knez, Ž. (2019) Chia Seeds (*Salvia Hispanica* L.): An Overview—Phytochemical Profile, Isolation Methods, and Application. *Molecules*, **25**, Article No. 11. https://doi.org/10.3390/molecules25010011

[23] Fernandes, S.S. and de las Mercedes Salas-Mellado, M. (2017) Addition of Chia Seed Mucilage for Reduction of Fat Content in Bread and Cakes. *Food Chemistry*, **227**, 237-244. https://doi.org/10.1016/j.foodchem.2017.01.075

[24] Ullah, R., Nadeem, M., Khalique, A., Imran, M., Mehmood, S., Javid, A. and Hussain, J. (2016) Nutritional and Therapeutic Perspectives of Chia (*Salvia hispanica* L.): A Review. *Journal of Food Science and Technology*, **53**, 1750-1758. https://doi.org/10.1007/s13197-015-1967-0

[25] Ixtaina, V.Y., Mattea, F., Cardarelli, D.A., Mattea, M.A., Nolasco, S.M. and Tomás, M.C. (2011) Supercritical Carbon Dioxide Extraction and Characterization of Argentinean Chia Seed Oil. *Journal of the American Oil Chemists' Society*, **88**, 289-298. https://doi.org/10.1007/s11746-010-1670-2

[26] Bochicchio, R., Philips, T.D., Lovelli, S., Labella, R., Galgano, F., di Marisco, A., Perniola, M. and Amato, M. (2015) Innovative Crop Productio*ns for Healthy Food: The Case of Chia (Salvia hispanica L.)*. In: Vastola, A., Ed., *The Sustainability of Agro-Food and Natural Resource Systems in the Mediterranean Basin*, Springer, Cham, 29-45. https://doi.org/10.1007/978-3-319-16357-4_3

[27] Akkarachaneeyakorn, S. and Tinrat, S. (2015) Effects of Types and Amounts of Stabilizers on Physical and Sensory Characteristics of Cloudy Ready-to-Drink Mulberry Fruit juice. *Food Science & Nutrition*, **3**, 213-220. https://doi.org/10.1002/fsn3.206

[28] Ikeda, S., Nitta, Y., Kim, B.S., Temsiripong, T., Pongsawatmanit, R. and Nishinari, K. (2004) Single-Phase Mixed Gels of Xyloglucan and Gellan. *Food Hydrocolloids*, **18**, 669-675. https://doi.org/10.1016/j.foodhyd.2003.11.005

[29] Katzbauer, B. (1998) Properties and Applications of Xanthan Gum. *Polymer Degradation and Stability*, **59**, 81-84. https://doi.org/10.1016/S0141-3910(97)00180-8

[30] Sworn, G. (2009) Gellan Gum. In: Phillips, G.O. and Williams, P.A., Eds., *Handbook of Hydrocolloids*, Vol. 37, Woodhead Publishing, Sawston, 204-227. https://doi.org/10.1533/9781845695873.204

[31] Leal, A.R., Holanda, L.E.O., Soares, F.C.M., da Costa, J.N., Nascimento, L.G.L., do Carmo, J.S., da Silva, W.C., Marques, L.F. and de Sousa, P.H.M. (2022) Effect of Gellan Gum Concentration on the Physicochemical, Rheological and Sensory Properties of Acerola Smoothie. *Food Science and Technology*, **42**, e05721. https://doi.org/10.1590/fst.05721

[32] Zhao, L., Pan, F., Mehmood, A., Zhang, Y., Hao, S., Rehman, A.U., Li, J., Wang, C. and Wang, Y. (2020) Protective Effect and Mechanism of Action of Xanthan Gum on the Color Stability of Black Rice Anthocyanins in Model Beverage Systems. *International Journal of Biological Macromolecules*, **164**, 3800-3807. https://doi.org/10.1016/j.ijbiomac.2020.09.027

[33] Hajmohammadi, A., Pirouzifard, M., Shahedi, M. and Alizadeh, M. (2016) Enrichment of a Fruit-Based Beverage in Dietary Fiber Using Basil Seed: Effect of Carboxymethyl Cellulose and Gum Tragacanth on Stability. *LWT-Food Science and Technology*, **74**, 84-91. https://doi.org/10.1016/j.lwt.2016.07.033

[34] Younes, M., Aggett, P., Aguilar, F., Crebelli, R., Filipic, M., Frutos, M. J., Galtier, P., Gott, D., Gundert-Remy, U., Kuhnle, G.G., Lambré, C., Leblanc, J., Lillegaard, I.T., Moldeus, P., Mortensen, A., Oskarsson, A., Stankovic, I., Waalkens-Berendsen, I., Woutersen, R.A. and Dusemund, B. (2018) Re-Evaluation of Gellan Gum (E 418) as Food Additive. *EFSA Journal*, **16**, e05296. https://doi.org/10.2903/j.efsa.2018.5296

[35] Cano-Sarmiento, C., Téllez-Medina, D.I., Viveros-Contreras, R., Cornejo-Mazón, M., Figueroa-Hernández, C.Y., García-Armenta, E., Alamilla-Beltrán, L., García, H.S. and Gutiérrez-López, G.F. (2018) Zeta Potential of Food Matrices. *Food Engineering Reviews*, **10**, 113-138. https://doi.org/10.1007/s12393-018-9176-z

[36] Xu, D., Zhang, J., Cao, Y., Wang, J. and Xiao, J. (2016) Influence of Microcrystalline Cellulose on the Microrheological Property and Freeze-Thaw Stability of Soybean Protein Hydrolysate Stabilized Curcumin Emulsion. *LWT-Food Science and Technology*, **66**, 590-597. https://doi.org/10.1016/j.lwt.2015.11.002

[37] Azarikia, F. and Abbasi, S. (2010) On the Stabilization Mechanism of Doogh (Iranian Yoghurt Drink) by Gum Tragacanth. *Food Hydrocolloids*, **24**, 358-363. https://doi.org/10.1016/j.foodhyd.2009.11.001

[38] Timilsena, Y.P., Wang, B., Adhikari, R. and Adhikari, B. (2016) Preparation and Characterization of Chia Seed Protein Isolate-Chia Seed Gum Complex Coacervates. *Food Hydrocolloids*, **52**, 554-563. https://doi.org/10.1016/j.foodhyd.2015.07.033

[39] Liang, C., Hu, X., Ni, Y., Wu, J., Chen, F. and Liao, X. (2006) Effect of Hydrocolloids on Pulp Sediment, White Sediment, Turbidity and Viscosity of Reconstituted Carrot Juice. *Food Hydrocolloids*, **20**, 1190-1197. https://doi.org/10.1016/j.foodhyd.2006.01.010

[40] Bhattacharjee, S. (2016) DLS and Zeta Potential—What They Are and What They Are Not. *Journal of Controlled Release*, **235**, 337-351. https://doi.org/10.1016/j.jconrel.2016.06.017